SELF FLOWER-COLOR INHERITANCE AND MUTATION IN MIRABILIS JALAPA L.

HIRAM M. SHOWALTER
Blandy Experimental Farm, University of Virginia, University, Virginia

Received December 14, 1933

Mirabilis jalapa, commonly known as "Four O'clock," "Sweet Marvel of Peru," or "Printoponite," belongs to the Nyctaginaceae. This species is cultivated for the brilliant color and pleasing odor of its flowers. Due to its simplicity, the breeding behavior of the self flower colors has furnished classical material for illustrating the simple laws of inheritance. It may be shown, however, that the breeding behavior of these self flower colors is not so simple as was first implied, and that where formerly there were only a few flower-color classes recognized, now there are many. CORRENS (1902, 1904), the first to investigate this species genetically, failed in many cases to recognize and classify the flower colors correctly. His attention was chiefly directed toward an interpretation of the "yellow×white" varietal cross in which reds (red being used in a very general sense to cover a number of different shades) were obtained in the F_1 and F_2, but he failed to distinguish between the F_2 red flower-color types. The careful distinction between the color classes came with the work of MARRYAT (1909). Aided by the discovery of "recessive white" (MARRYAT'S W7), she was able to detect the genotypes of all the material used in her investigations. The "rose pink" homozygote and "light pink" heterozygote of KIERNAN and WHITE (1926) were probably the first true pinks known to genetic literature on this species. The pink so frequently referred to in text-book illustrations as resulting from a "red×white" four o'clock cross is probably not pink, but magenta (see also MARRYAT 1909, and KIERNAN and WHITE 1926).

M. jalapa was introduced into Europe by the Spaniards in 1596. It is native to Peru, as one of its common names, "Sweet Marvel of Peru," suggests. HEIMERL (1901) found it to be native also to northern Mexico and the southern boundary of the United States. Under cultivation color varieties not known to the wild condition have occurred, and are recurring, spontaneously.

MATERIALS AND METHODS

This investigation (1) reviews all crosses, and reciprocals, previously made with true-breeding self flower-color varieties of *M. jalapa;* (2) attempts to describe the breeding behavior of Flesh Pink and Light Rosaline Purple, two new true-breeding types which have been found during the progress of the investigation, and reports the occurrence of Violet-Purple and Light Buff; and (3) attempts to throw light on the nature of mutation in flower color in this species.

In many respects *M. jalapa* is desirable material for crossing work. Its pollen grains are large (about 189μ in diameter) and can be administered singly to the stigma if desired. It is self-fertilized, its flowering season is long, it sets seed profusely, any particular plant can be carried over from one year to another by roots, and the characters in general are clear cut. However, in many cases, the flower-color heterozygotes cannot be distinguished at sight, as was formerly thought. Undesirable characteristics may be mentioned, such as the setting of only one seed per flower, the opening of the flowers for only a short time each day, and the susceptibility to cross-pollination by bees and nocturnal moths. In order to insure self- or cross-pollination, as the case may be, frames covered with cheese cloth have been used satisfactorily. The mature buds anthese about 6 P.M. in July (at The Blandy Experimental Farm), or about one half hour before sunset, and remain open until shortly after sunrise the next morning. Depending on temperature conditions this is variable, as is also the time of dehiscence of the anthers. The flowers with the darker hues open first. In crossing it is necessary to emasculate the mature buds of the female parent about three hours before time of anthesis, thus eliminating the possibility of self-pollination by precocious dehiscence of its anthers. In unemasculated flowers the style and anther filaments coil about each other some hours after anthesis, and if cross-pollination has not already taken place, self-fertilization will occur from self-pollen placed upon the stigma by this coiling.

The true-breeding self-flower color varieties of *M. jalapa* known at the present are Crimson, Yellow, Dominant White, Recessive White, Rose Pink, Flesh Pink, and Light Rosaline Purple. To this list probably will be added Violet-Purple and Light Buff. These last two have not been studied sufficiently to ascertain their breeding behavior (they were only discovered in 1933). Light Rosaline Purple has been used in only a few crosses. The known homozygotes are given in table 2. Similarly in table 3 the heterozygotes are given, with their corresponding genotypes.

The names of the homozygotes and heterozygotes describe as accurately as possible the colors of the flowers, and are given on the basis of comparisons with Ridgway's Color Standards and Nomenclature (1912). MARRYAT (1909) has plates of Crimson, Magenta, Magenta Rose, Yellow, White, et cetera, classes. The calyx makes up the showy part of the flower, having a tubular throat from 4 to 5 cm in length, and a rotate margin from 3.5 to 4.5 cm across, the latter being the highly colored portion. The throat color, inside and out, plus a star-shaped region on the upper surface running from the throat to the margin, is less intensely colored than the margin (see text figure 1). In "red" flowers the throat color is dull magenta; in "yellows," greenish yellow; in "whites," greenish white; and in "pinks," greenish pink. The style and anther filaments are generally about the same color as the throat of the flower, but deepen in intensity toward the extremities. The anthers (5 in number) and stigma correspond in color to the throat, except that they are more intensely colored. The stalk internodes, and particularly the nodes, are always somewhat magenta-colored in plants with "red" flowers; but in all others the stalks are entirely green.

On the basis of related genotypes and flower colors there are nine groups of heterozygotes (table 3). It is obvious from a study of this table that in many cases, types within a group cannot readily be distinguished at sight.

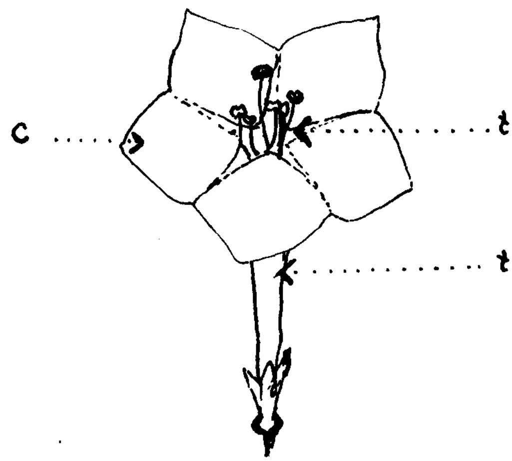

FIGURE 1.—Diagram of the color regions of the *M. jalapa* flower. c—region of intense color, t—color of the throat, less intense.

Depending on environmental conditions, such as sunlight, soil, et cetera, two plants of the same genotype may vary in intensity of color. A plant in the early flowering stages generally bears flowers which are a shade darker than those borne at a later stage. Due to this fluctuation in color, Yellow and Pale Yellow, Rose Pink and Light Pink, Light Rosaline Purple and Pale Rosaline Purple, homozygotes and heterozygotes respectively, may not be distinguishable. Naturally it is impossible to distinguish between the different genotypes of Crimson, Yellow and White, except by breeding.

Stocks of *M. jalapa* used in this investigation were grown from seed obtained from the following sources:—

(1) BROOKLYN BOTANIC GARDEN of the same lines used in the investigations of KIERNAN and WHITE (1926); and

(2) STARK BROTHERS, Seedsmen and Nurserymen, Louisiana, Missouri (Original Burbank stock).

FLOWER-COLOR INHERITANCE

At this point it is essential that a brief summary be made of the mechanism of flower-color inheritance. Crimson, Yellow, and Dominant White are the most common of the true-breeding varieties. In Yellow, there is a soluble yellow pigment base. ONSLOW (1925) thinks that all the varieties of *M. jalapa* "sprang originally from the crimson, which apparently contains a mixture of magenta anthocyanin and soluble yellow pigment." By the loss of anthocyanin the yellow variety was obtained, and by the loss of yellow pigment, the white varieties resulted.

The whole range of flower-color classes (see tables 2 and 3) of this species has been interpreted as being due to the action and interaction of factors at two loci (CORRENS 1902, 1904, MARRYAT 1909, KIERNAN and WHITE 1926). The one controls the color base, the other modifies the color base. Beginning with the work of KIERNAN and WHITE (1926) it may be shown that the latter locus is typified by an allelic series. The loci may be designated as the *Y* and *R* loci, respectively, using the terminology of KIERNAN and WHITE (1) with additional symbols to indicate the other factors of the *R* series (2).

Symbols:—

(1) *Y*—factor for yellow color, or color base;

y—allele of *Y*, absence of color base;

R—factor which modifies *Y* to red, and in the absence of *Y*, white color results;

Rp—allele of *R*, modifies *Y* to red, and when *Y* is replaced by *y*, it gives rose pink color;

r—allele of *R* and *Rp*, gives non-modification with *Y*, and in its absence (in the presence of *y*) white color results;

(2) *rp*—allele of the *R* series, gives non-modification with *Y*, and when *Y* is replaced by *y*, flesh pink color results. *rp* is dominant to *r*;

Rpl—factor for light rosaline purple, also allele of the *R* series. It is incompletely dominant to *R* and *Rp*, and presumably dominant to *r* and *rp*. In the presence of *Y* it produces red color. A complete account of its breeding behavior cannot be given at present.

For clearness the several crosses made during this investigation will be treated individually. Reciprocal crosses give the same results and are incorporated in table 1. From all the data obtained by myself, as well as those of previous experimenters, it is evident that the factors *Y* and *R* are inherited independently of each other.

(1) Crimson×Yellow. Three genotypes of Crimson have been identified in this investigation (see table 2-1, 2 and 3-1), which were postulated by KIERNAN and WHITE (1926). Those with genotypes *YYRR* and

YYRpRp breed true, while the third, *YYRRp*, upon selfing gives the first, second, and third types in the ratio of 1:1:2. On different occasions I have used the first and second types in this cross. Corresponding to the Crimson genotypes there are three of Yellow, *YYrr*, *YYrprp*, and *YYrrp* (see table 2-3, 4 and 3-2). The first and second breed true, while the third gives again the three types in an approximate 1:1:2 ratio. The F_1 heterozygote of this cross is Orange Red, Scarlet Red, or a shade intermediate to these colors, depending upon the parental genotypes. In the presence of *R*, orange red color is expected, and in the presence of *Rp*, scarlet red (see table 3-5, 6, 7, 8). The crosses from which the data are given (see table 1-1a-1b) had as the Crimson parents *YYRR*, and *YYRpRp* types, respectively. In the F_2 progeny, Crimson, Orange Red (or Scarlet Red), and Yellow, are obtained in an approximate 1:2:1 ratio.

(2) Crimson×Dominant White. Magenta is the only F_1 heterozygote which has been reported for this cross. By appropriate crosses, using Crimson and Dominant White, of the *YYRpRp* and *yyRR* types, respectively, Rhodamine Purple has been obtained (see table 1-2b). This heterozygote may appear indistinguishable from that of the Crimson×Rose Pink cross, but genotypically it is different, and theoretically less intense in color. By still further varying the cross, using a Crimson parent of the *YYRRp* type, two color heterozygotes should be obtained, Magenta and Rhodamine Purple. This cross has been made, but the results are not as satisfactory as theoretically they should be. Due to the similarity of these colors, it is extremely difficult to identify them in a population, even with the aid of a color chart. In the F_2, Crimson, Magenta (or Rhodamine Purple), and White (in the presence of the factor *Rp* in the homozygous condition, 1 White: 2 Light Pink: 1 Rose Pink, would be expected instead of all White) are obtained in an approximate 1:2:1 ratio.

(3) Crimson×Rose Pink. The original of this cross was not satisfactory, as pointed out by KIERNAN and WHITE (1926). But the postulation that Crimson, Rhodamine Purple, and Rose Pink would be obtained in an approximate 1:2:1 ratio in the F_2 has been verified in my work (see table 1-3). The F_1 heterozygote may be of the *YyRRp* or *YyRpRp* type (see table 3-14, 15), depending upon the genotype of the Crimson parent. I have not been successful in distinguishing between plants differing in this respect.

(4) Crimson×Flesh Pink. The Crimson of this cross was of the genotype, *YYRpRp*, and the Flesh Pink of the *yyrprp* type. The Rose Red of the F_1 is indistinguishable from that of the Yellow×Rose Pink cross, though theoretically it is of a deeper shade. It is of the genotype *YyRprp* (see table 3-12). The F_2 population is distributed in the approximate ratio of 1 Crimson:1 Yellow:3 Rose Pink:1 Flesh Pink:2 Scarlet Red:2 Rhoda-

TABLE 1

Data from crosses of flower-color varieties of M. jalapa.

CROSSES: PARENT GENOTYPE × PARENT GENOTYPE	F_2 COLORS	CRIMSON	YELLOW	WHITE	ROSE PINK	FLESH PINK	L. ROS. PURPL.	ORANGE RED	SCARLET RED	MAGENTA	RHOD. PURPLE	MAGENTA ROSE	ROSE RED	LIGHT PINK	PALE YELLOW	P. ROS. PURPL.	TOTALS
1a. Crimson *YYRR* × Yellow *YYrr*	ob.	40	42					87*									169
	ex.	42.2	42.2					84.5									
1b. Crimson *YYRpRp* × Yellow *YYrr*	ob.	26	31						65*								122
	ex.	30.5	30.5						61.0								
2a. Crimson *YYRR* × Dom. White *yyRR*	ob.	24		30						65*							119
	ex.	29.7		29.7						59.5							
2b. Crimson *YYRpRp* × Dom. White *yyRR*	ob.	13		17							52*						82
	ex.	20.5		20.5							41.0						
3. Crimson *YYRR* × Ro. Pink *yyRpRp*	ob.	32			42						73*						147
	ex.	36.7			36.7						73.5						
4. Crimson *YYRpRp* × Fl. Pink *yyrprp*	ob.	9	10		19	7			16		20		28*		17		126
	ex.	7.8	7.8		23.6	7.8			15.7		15.7		31.4		15.7		
5. Crimson *YYRpRp* × Rec. White *yyrr*	ob.	5	2	7	6				11		13		21*	16	14		95
	ex.	5.9	5.9	5.9	5.9				11.8		11.8		23.7	11.8	11.8		
6. Yellow *YYrr* × Dom. White *yyRR*	ob.	12	13	50				26		22		55*			26		204
	ex.	12.7	12.7	51				25.5		25.5		51.0			25.5		
7. Yellow *YYrprp* × Ro. Pink *yyRpRp*	ob.	20	24		20	24			41		40		67*	25	42		303
	ex.	18.9	18.9		18.9	18.9			37.8		37.8		75.7	37.8	37.8		
8. Yellow *YYrprp* × Fl. Pink *yyrprp*	ob.		24			32									36*		92
	ex.		23.0			23.0									46.0		
9. Yellow *YYrr* × Rec. White *yyrr*	ob.		13	9											24*		46
	ex.		11.5	11.5											23.0		
10. Dom. White *yyRR* × Ro. Pink *yyRpRp*	ob.			29	22									48*			99
	ex.			24.7	24.7									49.5			
11. Dom. White *yyRR* × Fl. Pink *yyrprp*	ob.			2		4								7*			13
	ex.			3.2		3.2								6.5			
12. Dom. White *yyRR* × Rec. White *yyrr*	ob.			17*													17
	ex.			17													
13. Ro. Pink *yyRpRp* × Fl. Pink *yyrprp*	ob.				53*	20											73
	ex.				54.6	18.2											
14. Ro. Pink *yyRpRp* × Rec. White *yyrr*	ob.			12	8									26*			46
	ex.			11.5	11.5									23.0			
15. Fl. Pink *yyrprp* × Rec. White *yyrr*	ob.			7		27*											34
	ex.			8.5		25.5											
16. L. Ros. Pur. *yyRplRpl* × Dom. White *yyRR*	ob.			4			3									6*	13
	ex.			3.2			3.2									6.5	

* F_1 flower color of respective crosses.

mine Purple:2 Pale Yellow:4 Rose Red. No Whites were obtained in my results (see table 1–4), which would have been expected had the Crimson parent been of the *YYRR* type.

TABLE 2

Known flower-color homozygotes of M. jalapa. Light Rosaline Purple is excepted since its genotypic formula is uncertain.

(1) *YYRR* Crimson	(3) *YYrr* Yellow
(2) *YYRpRp* Crimson	(4) *YYrprp* Yellow
(5) *yyRR* Dominant White	(7) *yyRpRp* Rose Pink
(6) *yyrr* Recessive White	(8) *yyrprp* Flesh Pink

TABLE 3

Groups representing related genotypes of flower-color heterozygotes of M. jalapa.

(1) *YYRRp* Crimson (group 1)	(2) *YYrrp* Yellow (group 2)
(3) *yyRr* White (group 3)	(4) *yyrrp* Flesh Pink (group 4)
(5) *YYRr*	(9) *YyRr*
(6) *YYRpr* Orange Red to Scarlet Red	(10) *YyRpr* Magenta Rose to Rose Red (group 6)
(7) *YYRrp* (group 5)	(11) *YyRrp*
(8) *YYRprp*	(12) *YyRprp*
(13) *YyRR*	(16) *Yyrr*
(14) *YyRRp* Magenta to Rhodam. Purp.	(17) *Yyrrp* Pale Yellow (group 8)
(15) *YyRpRp* (group 7)	(18) *Yyrprp*
(19) *yyRRp*	
(20) *yyRpr* Light Pink to Rose Pink (group 9)	
(21) *yyRrp*	
(22) *yyRprp*	

(5) Crimson×Recessive White. Difficulty was encountered in obtaining the Recessive White for this cross (see table 2–6). This type was originally discovered by MARRYAT (1909), and is distinct from the Dominant White. In the F_1 of this cross MARRYAT obtained Magenta Rose plants, indistinguishable from that of the Yellow×Dominant White cross. In my experience I have obtained Rose Red (see table 3–10) in the F_1, due to the *Rp* factor of the Crimson parent, not present in MARRYAT's experiments. In the F_2 approximately 1 Crimson:1 Yellow:1 White:1 Rose Pink:2 Orange Red to Scarlet Red:2 Rhodamine Purple:2 Light Pink:2 Pale Yellow:4 Rose Red occurred (see table 1–5). Had factor *Rp* not been present in the Crimson parent, more Whites would have been expected.

(6) Yellow×Dominant White. Magenta Rose was obtained in the F_1 (see table 3–9), the parental genotypes being *YYrr* and *yyRR*, respectively. No "flaking" occurred in a total of 19 plants. "Flaking" is the term used by MARRYAT (1909) to designate bi- and tri-colored flowers in *M. jalapa;* for example, flowers marked with crimson and white, yellow and white, or crimson, yellow and white, et cetera. The color markings in bi- and tri-colored forms vary, ranging from a few small dots or striations to an area half or three-quarters of the flower. According to MARRYAT "it is characteristic of Mirabilis that bi- and tri-coloured individuals almost always produce a few whole-coloured flowers; *e.g.* a plant having most of its flowers white flaked with yellow, bears some pure yellow and some pure white blossoms, and so on.") MARRYAT (1909) found flaking in all her F_1 plants of this cross and thought it was peculiar to the particular cross, whereas in my experience it is due rather to the material. Her material must not have been homozygous for self-flower color. My results of the F_2 population of this cross (see table 1–6) show a close dihybrid ratio of 1 Crimson:1 Yellow:4 White:2 Orange Red:2 Magenta:2 Pale Yellow:4 Magenta Rose. F_3 and F_4 generations of this cross have been grown to check upon the occurrence of flaking. Flower-color mutations were observed, but not in excess of what would be expected of any *M. jalapa* material.

(7) Yellow×Rose Pink. This cross gave results similar to the original of KIERNAN and WHITE (1926), except that in my work, Yellow of the *YYrprp* type was used instead of the *YYrr* of the former work. These Yellows are indistinguishable, except in breeding experiments. The F_1 Rose Red (see table 3–12) gave an F_2 of Crimson, Yellow, Rose Pink, Flesh Pink, Scarlet Red, Rhodamine Purple, Pale Yellow, Light Pink and Rose Red; in an approximate 1:1:1:1:2:2:2:2:4 ratio (see table 1–7).

(8) Yellow×Flesh Pink. The F_1 heterozygote of this cross is Pale Yellow (see table 3–17 or 18), indistinguishable from that of the Yellow× Dominant White cross, though theoretically it shows more pink. In the F_2, Yellow, Pale Yellow, and Flesh Pink were obtained closely approximating a 1:2:1 ratio. The parents in my crosses had the genotypes *YY rp rp* and *yy rp rp*, respectively. As a proof of this, no Whites occurred in the F_2. White would be expected when either parent carries the factor *r* instead of *rp*.

(9) Yellow×Recessive White. In the F_2 an approximate 1:2:1 ratio of Yellows, Pale Yellows, and Whites was obtained (see table 1–9). The Yellow parent was derived from a strain homozygous for *r* (see table 2–3). This cross was used to check on the genotypes of the Yellows.

(10) Dominant White×Rose Pink. The results are similar to those of

KIERNAN and WHITE (1926). The Light Pink heterozygote (see table 3–19) is difficult, or impossible, to distinguish from those of the Dominant White×Flesh Pink, and Rose Pink×Recessive White crosses (see table 1–11, 14). The results of this cross are given in table 1–10.

(11) Dominant White×Flesh Pink. This cross gave an F_1 Light Pink (see table 3–21), probably less intense than that from the Dominant White ×Rose Pink cross. In the F_2, Whites, Light Pinks, and Flesh Pinks are obtained in an approximate 1:2:1 ratio (see table 1–11).

(12) Dominant White×Recessive White. As expected, this cross gave only White in the F_1 and F_2 (see table 1–12). For the genotypes of White see tables 2–5, 6 and 3–3.

(13) Rose Pink×Flesh Pink. The F_1 heterozygote, Rose Pink, is only slightly, if any, lighter than the Rose Pink parent (see table 3–22 and 2–7, respectively). In the F_2 Rose Pink and Flesh Pink were obtained closely approximating a 3:1 ratio (see table 1–13), showing that factor Rp for Rose Pink is dominant over rp, the factor for Flesh Pink.

(14) Rose Pink×Recessive White. The F_1 heterozygote of this cross is difficult, or impossible, to distinguish from the Light Pink of other crosses, as for instance the cross, Dominant White×Rose Pink. Upon selfing, however, it gives a close 1:2:1 ratio of Rose Pink, Light Pink, and White (see table 1–14).

(15) Flesh Pink×Recessive White. Flesh Pink is obtained in the F_1, indicating that the factor rp for Flesh Pink is dominant over r, the factor for Recessive White. Only a few F_2 plants have been grown from this cross, but of these a somewhat poor approximation to a 3:1 ratio of Flesh Pink and White was obtained (see table 1–15). Some flaking occurred in the F_1 and F_2 progeny of this cross.

(16) Dominant White×Light Rosaline Purple. This is the only cross involving Light Rosaline Purple which has yielded an F_2 to date. In the F_1 a purple lighter than the purple parent is obtained. It is analogous to the heterozygous stock from which Light Rosaline Purple was first obtained, namely: Pale Rosaline Purple. Upon selfing, the F_1 heterozygote gives an F_2 of Light Rosaline Purple, Pale Rosaline Purple, and White, in approximately a 1:2:1 ratio (see table 1–16).

The behavior of Light Rosaline Purple indicates that it carries a factor allelic to the R series. It breeds true, and on the basis of the observations already made on this new flower type, the factor symbol Rpl is given it. The F_1 of a cross, Crimson×Light Rosaline Purple, is of a darker shade of Rhodamine Purple than that of the cross, Crimson×Rose Pink. In the cross, Rose Pink×Light Rosaline Purple, Pale Rosaline Purple, apparently indistinguishable from that of the Dominant White×Light Rosaline Purple, is obtained. The factor Rpl gives more color, both in the presence and in the absence of Y, than does Rp, or any other factors of the R series. It is practically dominant over the factors Rp and R, and probably should be listed at the top of the R series.

FLOWER-COLOR MUTATION

It is not uncommon to see a plant of *M. jalapa* bearing two distinctly different flower colors, in the same, or in different flowers. In cases where mutation occurs in self-colored plants, there is generally a preponderance of flowers of the same color, with a few of a different, or mutant color. These few are borne on one (rarely more than one) terminal branch, indicating that somatic mutation took place late in the growth of the plant, giving a new flower color. In some cases not all the flowers of such a branch are self-colored. A single blossom may show part mutant and part non-mutant color, indicating that the whole of the growing tip of the branch was not involved in the mutation. This suggests the problem of "flaking" to which MARRYAT (1909) and CORRENS (1910) gave so much of their attention while working with this species. The phenomenon will not be discussed further in this paper.

Of the 5,000 plants (slightly more) of *M. jalapa* grown since this investigation began, 21 self flower-color mutations have been observed and recorded. Most of the observations have been confined to material with known breeding behavior.

The somatic mutations observed are as follows:

(1) White with Light Pink branch (R to Rp) 6 cases
(2) White with Light Pink branch (R to rp) 2 cases
(3) White with Pale Rosaline Purple branch (R to Rpl) 3 cases
(4) White with Light Buff branch (? to ?) 3 cases
(5) Rose Pink with Light Pink branch (Rp to R) 1 case
(6) Crimson with Orange Red branch (R to r) 4 cases
(7) Flesh Pink with Light Pink branch (rp to R) 2 cases

It is very probable that Flesh Pink mutates to Rose Pink (rp to Rp), and that Dominant White and Flesh Pink both mutate to Recessive White (R and rp to r), but that in the latter two cases the mutation is covered by dominancy. It is probable that the total number of somatic mutations in self-flower color is appreciably higher than the observed number (21 per 5,000 individuals), probably one mutation per sixty individuals. It should be stated that the observed mutation frequency is noticeably higher in some flower-color classes than in others; and it may be higher in one line than in another of the same genotype. With one exception I have found the mutant characters in the heterozygous condition. In that case I obtained four seeds from a branch bearing presumably Rose Pink flowers on a plant whose other flowers were White. The three plants obtained from these seed were classified as Rose Pink. If this be true, it would mean that double mutation (involving genes in homologous chromosomes) had taken place. An F_3 of these plants may reveal even yet, that this was a single mutation, and that a Light Pink mutation was mistaken for Rose Pink.

In addition to somatic mutation, a few cases of what appears to be gametic mutation have been found. Pale Rosaline Purple (1 plant) was obtained in the F_2 population of a cross, Crimson (*YY Rp Rp* type) $\times$ Dominant White, out of a total of 90 plants (in 1931). Another case of the same kind occurred in 1933 in an F_2 population of a cross, Dominant White $\times$ Flesh Pink, giving Violet-Purple, the only instance of this color class. Just what this type will give upon selfing is not known, but like Pale Rosaline Purple, it will probably give rise to a new true-breeding color variety, analogous to Light Rosaline Purple.

DISCUSSION

Prior to the discovery of the factor *rp*, it was comparatively easy to identify the individuals in a hybrid population of *M. jalapa* plants as to their genotypes, except for the Whites. Since its discovery it is difficult, and in most cases impossible, to distinguish the flower-color classes, except by breeding tests. Yellow, formerly thought to be of only one genotype, is known to have three. So far as I am concerned they are not distinguishable, though theoretically they should be. Those having the factor *rp* in their complex should show a trace of pink, not expected in the original Yellow with *r* instead of *rp*. Similarly in Crimson there are three known genotypes (and probably more will be had), depending on the factors of the *R* series present, whether *R* itself, or its allele *Rp*, in the homozygous condition; or both in the heterozygous condition. Each has a distinct action in modifying *Y* (factor for color base) to red, but I find that the phenotypic expression from one type to another, is not sufficiently affected to distinguish between them accurately. With the discovery of the factor for Light Rosaline Purple, tentatively designated *Rpl*, which also possesses the power to modify *Y* to red, a third homozygous Crimson type is postulated, which theoretically will show more purple color than those already known, but actually it may appear indistinguishable. Its behavior in the cross, Crimson (of the *YYRpRp* type) $\times$ Light Rosaline Purple, giving the heterozygote, Rhodamine Purple, darker than that of the Crimson $\times$ Rose Pink cross, indicates that the factor *Rpl* may show up distinctly in Crimson where reddening is at its maximum. This would bring the total of Crimson genotypes up to six, and would increase the number of heterozygotes with Crimson proportionally. As in Crimson and Yellow, there is more than one type recognized of Rose Pink, Flesh Pink, and White.

Factors R, Rp, and Rpl all produce reddening in the presence of Y. In addition to reddening, Rp produces pinking and slight purpling. In addition to reddening, pinking and slight purpling, Rpl produces deeper purpling, thus combining and exceeding the action of the first two, R and Rp. This justifies naming it at the top of the R series, with Rp and R next in order. rp like r does not produce reddening in the presence of Y, but it is dominant to r, as revealed in the cross, Flesh Pink×Recessive White. In turn rp is recessive to Rp, or nearly so as seen from the cross, Rose Pink×Flesh Pink. On the basis of their behavior, the alleles of the R series may be arranged as follows, beginning at the top: Rpl, Rp, R, rp, and r. Upon further investigation of the new self-color types, Violet-Purple and Light Buff, the series may be lengthened.

No evidence has been accumulated to show that any of the alleles of the R series are stable. No observations have been made of r changing to rp, or rp to Rp, but since rp is dominant to r, and Rp to rp, these reverses could not have been seen at once, and would be lost in the population. Of the mutations in self flower-color so far observed, 14 represent changes from R to some allele, except perhaps in the case where Violet-Purple occurred. Rp stands next in the number of changes observed with 5; and rp third with 2. The individuals in my populations having Rp and rp in their complex have been as numerous as those with R, but have not shown as great a number of changes. Rpl, factor for Light Rosaline Purple, has arisen from R three times, but not once from Rp to date. Light Buff has arisen from White plants.

KIERNAN and WHITE's postulation (1926) that the factor Rp arose by mutation from R has been verified in this investigation. At least 6 cases have been observed where White Plants have born Light Pink flowers. Seeds obtained from these branches yield Rose Pink, Light Pink, and White plants. Rp and R have both reverted in my material to rp for Flesh Pink. Since practically all of my material was grown from stocks obtained from KIERNAN and WHITE, they must have had the same phenomenon too. Of the seed which I obtained from them, one package gave Flesh Pink plants. It was, however, labeled "Light Pink," indicating that they had Flesh Pink in their stocks, but that they did not distinguish it from Light Pink.

SUMMARY

1. Crosses and reciprocal crosses have been made of all the known true-breeding color varieties of *M. jalapa*, namely: Crimson, Yellow, Dominant White, Rose Pink, Flesh Pink, Recessive White, and Light Rosaline Purple. The latter has not been investigated thoroughly.

2. Data are presented on the spontaneous occurrence of varieties, Rose Pink, Flesh Pink, Light Rosaline Purple, Violet-Purple, and Light Buff.

The breeding behavior of Flesh Pink is given in detail, while Light Rosaline Purple, Violet-Purple, and Light Buff have yet to be investigated more thoroughly.

3. The whole range of flower-color classes is interpreted as being due to the action and interaction of factors at two loci, a *Y* (color base) and an *R* (modifying) locus.

4. The factors for Flesh Pink and Light Rosaline Purple have been isolated and designated *rp* and *Rpl*, respectively. These factors have been found to be alleles of the *R* series.

5. The alleles of the *R* series have been arranged as follows, beginning at the top: *Rpl*, *Rp*, *R*, *rp*, and *r*.

6. In the presence of *Y*, *Rpl* covers the action of *R* by producing reddening, and that of *Rp* by producing reddening, pinking, and slight purpling, and exceeds their action by giving more intense purpling.

8. *rp*, factor for Flesh Pink, is recessive to *Rp*, produces Light Pink in the presence of *R*, and is dominant to *r*. It does not possess the power to modify *Y* to red.

9. Factor *R* is probably less stable than any of its alleles.

10. Somatic mutation is common, taking place in the late ontogeny of the plant. Cases of gametic mutation have been observed, it is thought, but they are more rare.

11. The discovery of factor *rp* gives further complications to recognizing the various color classes in a population.

12. On the basis of related genotypes, 9 heterozygous groups have been recognized.

ACKNOWLEDGMENT

The writer is indebted to Dr. ORLAND E. WHITE for numerous suggestions and criticisms given during this study.

LITERATURE CITED

CORRENS, C., 1902 Ueber Bastardirungsversuche mit Mirabilis-Sippen. Ber. deutsch. bot. Ges. **20**: 504–608.

1904 Zur Kenntnis der scheinbar neuen Merkmal der Bastarde. Ber. deutsch. bot. Ges. **23**: 70–85.

1910 Der Übergang aus dem homozygotischen in einen heterozygotischen Zustand im selben Individuum bei buntblättrigen und gestreiftblühenden Mirabilis-Sippen. Ber. deutsch. bot. Ges. **28**: 418–434.

HEIMERL, A., 1901 Studien uber einige Nyctaginaceen des Herbarium Delessert. Annu. Conserv. Jard. bot. Genève. **5**: 178–179, 617.

KIERNAN, F. P. and WHITE, O. E., 1926 Inheritance in four o'clocks. J. Hered. **17**: 383–386.

MARRYAT, DOROTHEA C. E., 1909 Hybridization experiments with *Mirabilis jalapa*. Reports to the Evolution Committee of the Royal Society of London. **5**: 32–50.

ONSLOW, M. (Wheldale), 1925 The anthocyanin pigments of plants. Cambridge Univ. Press, Sec. Ed. : 151.

RIDGWAY, ROBERT, 1912 Color standards and nomenclature. Washington.